CLAIMS

The invention claimed is:

1. A method of isolating mutant yeast cells with an increased life span, comprising the steps of:

a) exposing a sample of yeast cells from a budding yeast strain, for which the life span is known, to a mutagen;

b) plating the sample of cells on minimal medium necessary for growth of yeast cells, thereby generating an original plate;

c) replica-plating the original plate to a plate with a medium lacking nutrients necessary for growth of yeast cells, thereby generating a replica plate;

d) culturing the original plate and the replica plate under conditions appropriate for growth of yeast cells;

e) replica-plating the replica plate to enriched medium, thereby generating an enriched plate;

f) culturing the enriched plates under conditions for growth of yeast cells, thereby generating colonies of yeast cells which are starvation resistant;

g) calculating the life span of yeast cells in the starvation resistant colonies; and

h) selecting those yeast cells with a life span that is longer than the known life span of the yeast strain.

2. A method of isolating mutant yeast cells with an increased life span, comprising the steps of:

a) exposing a sample of yeast cells from a budding yeast strain, for which the life span is known, to a mutagen;

b) labelling the cell surface of the yeast cells with a fluorescent marker, thereby generating fluorescent yeast cells;

c) culturing the yeast cells under conditions for growth of yeast cells, and for a period of time greater than the chronological life span of the strain;

d) subjecting the yeast cells to fluorescence-activated cell sorting, thereby separating fluorescent yeast cells from non-fluorescent yeast cells;

e) replating the fluorescent yeast cells, under conditions for growth of yeast cells;

wherein those fluorescent yeast cells which grow after replating are mutant yeast cells having an increased life span.

3. A method of isolating mutant yeast cells with an increased life span, comprising the steps of:

a) exposing a sample of yeast cells from a temperature-sensitive budding yeast strain, in which the daughter cells die at the nonpermissive temperature, and for which the life span is known, to a mutagen;

b) plating the yeast cells, and cultivating the yeast cells at the permissive temperature and under conditions for growth of yeast cells;

c) transferring a sample of yeast cells from each colony of the plate at the permissive temperature to a second plate;

d) cultivating the yeast cells transferred to the second plate at the nonpermissive temperature, thereby generating microcolonies of yeast cells; and

e) calculating the number of yeast cells in the microcolonies,

wherein if a microcolony consists of a number of yeast cells that is greater than the number of generations in the life span of the yeast strain, the microcolony contains mutant yeast cells having an increased life span.

4. A method of isolating mutant yeast cells with an increased life span, comprising the steps of:

a) exposing a sample of yeast cells from a temperature-sensitive budding yeast strain, in which the daughter cells die at the nonpermissive temperature, and for which the life span is known, to a mutagen;

b) plating the yeast cells, and cultivating the yeast cells at the nonpermissive temperature for a period of time greater than the chronological life span of the strain, thereby generating microcolonies of yeast cells; and

c) shifting the microcolonies to the permissive temperature,

wherein those yeast cells which grow after the shift to the permissive temperature are mutant yeast cells having an increased life span.

5. An agent which increases the life span of yeast cells, isolated by the method of:

a) exposing a sample of yeast cells from a budding yeast strain, for which the life span is known, to the agent to be tested;

b) plating the sample of yeast cells with the minimal medium necessary for growth of yeast cells, thereby generating an original plate;

c) replica-plating the original plate to a plate with a medium lacking nutrients necessary for growth of yeast cells, thereby generating a replica plate;

d) culturing the original plate and the replica plate under conditions appropriate for growth of yeast cells;

e) replica-plating the replica plate to an enriched medium, thereby generating an enriched plate;

f) culturing the enriched plates under conditions for growth of yeast cells; and

g) calculating the life span of yeast cells which grow on enriched plates,

wherein the presence of yeast cells with a longer life span than that of the known life span of the yeast strain is indicative of the ability of the agent to increase life span.

6. An agent which increases the life span of yeast cells, isolated by the method of:

a) exposing a sample of yeast cells from a budding yeast strain, for which the life span is known, to the agent to be tested;

b) labelling the cell surface of the yeast cells with a fluorescent marker, thereby generating fluorescent yeast cells;

c) culturing the yeast cells under conditions for growth of yeast cells, and for a period of time greater than the chronological life span of the strain;

d) subjecting the yeast cells to fluorescence-activated cell sorting, thereby separating fluorescent yeast cells from non-fluorescent yeast cells;

e) replating the fluorescent yeast cells, under conditions for growth of yeast cells;

wherein growth of fluorescent yeast cells after replating is indicative of the capability of the agent to increase life span.

7. An agent which increases the life span of yeast cells, isolated by the method of:

a) exposing a sample of yeast cells from a temperature-sensitive budding yeast strain, in which the daughter cells die at the nonpermissive temperature, and for which the life span is known, to the agent to be tested;

b) plating the yeast cells, and cultivating the yeast cells at the permissive temperature and under conditions for growth of yeast cells;

c) transferring a sample of yeast cells from each colony of the plate at the permissive temperature to a second plate;

d) cultivating the yeast cells transferred to the second plate at the nonpermissive temperature, thereby generating microcolonies of yeast cells; and

e) calculating the number of yeast cells in the microcolonies,

wherein the existence of microcolonies consisting of a number of yeast cells that is greater than the number of generations in the life span of the yeast strain, is indicative of the capability of the agent to increase life span.

8. An agent which increases the life span of yeast cells, isolated by the method of:

a) exposing a sample of yeast cells from a temperature-sensitive budding yeast strain, in which the daughter cells die at the nonpermissive temperature, and for which the life span is known, to the agent to be tested;

b) plating the yeast cells, and cultivating the yeast cells at the nonpermissive temperature for a period of time greater than the chronological life span of the strain, thereby generating microcolonies of yeast cells; and

c) shifting the microcolonies to the permissive temperature,

wherein the growth of yeast cells after the shift to the permissive temperature is indicative of the capability of the agent to increase life span.

9. A protein that contributes to senescence in an organism encoded by a gene, isolated by a method comprising the steps of:

a) generating a genomic DNA library from the organism of interest;

b) transforming yeast cells from a budding yeast strain, in which the SIR4 gene has been mutated to generate a stop at codon 1237, with the library;

c) plating the transformed yeast cells, and culturing them under conditions for growth of yeast cells;

d) calculating the life span of the colonies of transformed yeast cells;

e) selecting yeast cells from colonies in which the life span of the yeast cells is approximately equal to the life span of the yeast strain in which the SIR4 gene has not been mutated to generate a stop at codon 1237, thereby obtaining yeast cells containing senescence genes from the organism of interest; and

f) isolating from the selected yeast cells the DNA from the organism of interest,

thereby obtaining a gene encoding a protein that contributes to senescence.

10. A protein that contributes to senescence in an organism, encoded by a gene which is homologous and/or hybridizes under conditions of medium stringency to a gene isolated by a method comprising the steps of:

a) generating a genomic DNA library from the organism of interest;

b) transforming yeast cells from a budding yeast strain, in which the SIR4 gene has been mutated to generate a stop at codon 1237, with the library;

c) plating the transformed yeast cells, and culturing them under conditions for growth of yeast cells;

d) calculating the life span of the colonies of transformed yeast cells;

e) selecting yeast cells from colonies in which the life span of the yeast cells is approximately equal to the life span of the yeast strain in which the SIR4 gene has not been mutated to generate a stop at codon 1237, thereby obtaining yeast cells containing senescence genes from the organism of interest; and

f) isolating from the selected yeast cells the DNA from the organism of interest,

thereby obtaining a gene encoding a protein that contributes to senescence.

11. The protein encoded by a mutant SIR4 gene, consisting essentially of a SIR4 gene having a stop at codon 1237.

12. A protein comprising a region which is homologous to the SUN domain region, said SUN domain region consisting essentially of an amino acid sequence selected from the group consisting of:

a) amino acid 236 through amino acid 451 of SEQ ID NO. 2;

b) amino acid 123 through amino acid 338 of SEQ ID NO. 12; and

c) amino acid 211 through amino acid 426 of SEQ ID NO. 14.

ADD A1